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REPORT: THE ANALYSIS OF AIR PARTICULATES
AND LEACHATES FROM THE UPPER OTTAWA
STREET LANDFILL FOR MUTAGENICITY IN THE
AMES TEST

November 1982

P.O. BOX 198 KLEINBURG, ONTARIO, CANADA L0J 1C0

SUMMARY

Air particulate and leachate samples collected on or around the Upper Ottawa Street Landfill Site have been analysed for mutagenicity in the Ames assay.

In experiments with TA98 and TA1537, in no case did the mutagenicity of extracts from air particulates collected on the site exceed the mutagenicity of air particulates obtained at control locations around Hamilton. Although the highest value for mutagenicity using TA100 was obtained with an extract from particulates collected on the landfill, this result could not be repeated and may have been a false positive. Activity of other particulate extracts on TA100 was low.

Only one clearly positive result was found amongst the leachate samples analysed. A doubling of background reversion frequency was obtained using leachate from bore hole #4 and TA98.

INTRODUCTION

This report provides details of the analysis of leachate and air particulate samples collected on or around the Upper Ottawa Street Landfill Site for mutagenicity in the Ames assay.

MATERIALS & METHODS

Mutagenic activity was assayed using histidine auxotrophs of *S.typhimurium*. Strains TA98,TA100,TA1535,TA1537 and TA1538 were kindly provided by Dr. Bruce Ames, Berkeley, Calif. and were stored as frozen stocks at -70°C . The characteristics of these stocks are checked periodically (Histidine requirement,rfa character, R-factor,uvrB deletion: Appendix).

Bacterial cultures for air particulate studies were established by scraping a sterile wooden applicator stick across the surface of the frozen stock, innoculating either 5 or 20ml nutrient broth and shaking overnight at 37°C . Cultures for leachate studies were from a 1/100 dilution of an overnight culture shaken for 6hr at 37°C .

Air Particulate Samples

Air particulates were collected using "hi-vol" samplers, provided by the MoE, operating over a 24hr period from noon Aug. 11,1982 to noon Aug.12th. Filters were delivered to Mutatech Inc. in manilla envelopes and were stored in the dark at 4°C . Before extraction each filter was left at room temperature for 48hrs,then weighed. Particulate weights are shown in Table I. Filters were extracted according to the method of Katz et al 1978.

Each filter was folded and placed in a glass extraction thimble with a sintered glass disc at one end. This was then placed in a Soxhlet extractor and extracted for 17 hours with 200ml methanol. After extraction the thimble was allowed to cool to room temperature and rinsed with 25ml methanol; all washings were collected in the boiling flask and the contents of the flask filtered through a sintered glass funnel. Flask and funnel were then rinsed 3x with 7ml methanol and the entire extract reduced to about 2ml on a rotary evaporator in a 50ml pear-shaped flask. The water bath temperature was maintained at approx. 30°C. The remaining solvent was then transferred to a glass vial, the pear-shaped flask rinsed 3x and the final extract evaporated to dryness under a stream of nitrogen. The residue was stored in the dark in the refrigerator.

Ames Assay

Each extract was dissolved in DMSO and further diluted such that 0.1ml sample was added per plate. For each experiment an appropriate aliquot was removed from each sample vial and the remainder re-frozen and stored for further use. The Ames assay was carried out as described by Ames et al, 1975. 0.2ml S9 mix (Salamone et al, 1979) was used where appropriate. The same lot of S9 homogenate (Litton Bionetics, Maryland) was used throughout the experiments in these studies except for those (two) carried out after 011182.

Leachate Samples

Leachate samples were collected from 5 sites on or around the Upper Ottawa Street Landfill Site, namely bore holes 2 and 4, Collection Manhole, Redhill Creek upstream and downstream. In addition, a sample of Hamilton tap water was provided for comparison. The leachates were delivered to Mutatech on 3 Aug. 1982, and stored

in the refrigerator in the dark. Shortly after receipt, the pH of each sample was measured. An aliquot of each was then vacuum filtered, first through Whatman #2 filter paper then through a 0.45 μ membrane filter. The final filtrates were stored as above. Before use samples were diluted .75, .5 and .25x with sterile deionized water.

Ames Assay

Top agar was concentrated such that 0.7ml could be added per plate. To this was added 2ml of sample, 0.1ml cells, and, where appropriate 0.2ml S.9 mix (Salamone et al 1979). 2ml deionized water was used as a negative control. Appropriate positive controls were included in addition to (in most experiments) control plates for S 9 mix and sample sterility.

RESULTS

Air particulates

The first mutagenicity/toxicity assays either on TA98 or TA100 were carried out with the intention of using all 5 tester strains. Therefore one fifth the total volume of each sample in solution in DMSO was removed and used on one or other of these strains. It was evident, however, that the maximum doses permitted by the limited quantity of material available were insufficient to cause toxicity and a change of protocol was proposed. TA1535 and 1538 were eliminated from the study allowing the maximum dose to be increased in subsequent experiments.

Any sample then remaining was to be used in a re-test of TA98 or TA100 as appropriate at a higher concentration.

In spite of the higher doses permitted, no toxicity, as indicated by the background bacterial lawn, was observed with any extract. The most mutagenic activity was observed with TA98 and, in most cases, this was enhanced by the presence of S 9 mix (Tables II-XI). In almost all experiments some evidence of dose response was observed. Linear regression analyses were carried out on the data and a value for revertants per mg particulate matter calculated for each sample from the linear part of each dose-response curve (Tables XII-XIV). From this it was possible to calculate revertants per m^3 air sampled and these data are shown in the same table. Although artificial, these values provide a convenient way of comparing extracts. With strains TA98 and TA1537 mutagenic activity per m^3 air was greatest in the sample taken at the MoE Instrumentation Lab at Stoney Creek, although this was clearly because particulate concentration was very high at this site (Table I). In terms of mutagenicity per mg particulate, no extract exceeded the mutagenicity of the control sample -001 (Mountain Police). On re-testing, the activities of the various extracts on TA98 were not inconsistent with initial results (Tables I-XI).

Only low levels of mutagenic activity were observed with most extracts on TA100 (Tables II-XI). The only sample with which a doubling of background reversion rate was obtained was -025 (North of Site, Near Gate). (Borehole #1)

This extract was more mutagenic both in terms of revertants/mg and revertants/m³ air than any other sample. When, however, the extract was re-tested on TA100, these levels of mutagenicity were not observed (Table XI). Extracts -002 (Blessed Kateri Tekakwitha School) and -004 (Bell Canada Building) were also re-tested on TA100 (Tables III and IV respectively). Results with -004 were in agreement with original observations of little or no mutagenicity. Although reduced mutagenic activity was observed with extract -002, this reduction was not so striking as that observed with -025. Reasons for the lack of reproducibility of results on re-testing of these samples on TA100 are unclear but four possible explanations are offered:

- (1). The outcome of the first Ames test of sample -025 and -002 (carried out at the same time, with the same controls) was a spurious positive due to technical error.
- (2) The outcome of the re-test was a false negative.
- (3) A new lot of S9 homogenate was used for the re-test.
- (4) The activity present in the samples declined during the 20 day period between the two experiments in spite of their being stored frozen.

The second explanation need not be considered because a third repeat (data not shown) using what remained of the sample from the final dilutions from each of the previous experiments (at 0.4ml/plate), again indicated low levels of reversion. It is not easy to dismiss any of the remaining 3 possibilities without further study.

Leachates

A doubling of background reversion rate and a clear indication of dose dependancy was shown using the sample from bore hole #4 and TA98, in both the presence and absence of S9 (Table XXI). A weak positive was also indicated with this sample and TA1538 without S9 (Table XXI). Similarly weak responses were observed in the absence of S9 with TA1538 and leachate from bore hole #2 (Table XVII) and with TA100 and the upstream sample from Redhill Creek (Table XVIII). That these increases were not the result of sample contamination was verified in each case either at the time of the experiment, by incorporation of sample into top agar, or subsequently, by shaking a 2ml aliquot with nutrient broth at 37°C.

REFERENCES

1. Ames, B.N., McCann, J., Yamasaki, E. (1975) Mutation Res.
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3. Salamone, M.F., Heddle, J.A., Katz, M. (1979) Environment Int.
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TABLE 1 COLLECTION DATA FOR AIRBORN PARTICULATES

LOCATION	Hi-vol #	Filter #	Collection Period(hrs)	Particulate Weight(mg)	Conc. Airborn Particulates ($\mu\text{g m}^{-3}$)
Instrumentation Laboratory (2284 Barton St.)	8893	-006	24	193.7	73.9
Site, East Side Borehole #4	8898	-008	24	109.5	41.6
Site, South Near Stonechurch Rd. (goose neck vents)	9495	-009	24	101.3	37.2
Bell Canada Building (on south side of Stonechurch)	9896	-004	24	97.1	36.7
Benneto School (Huguenot St. W.) across from damp	9739	-005	24	89.7	31.9
Site, South Side - Borehole #3	9258	-007	18	65.4	30.9
Mountain Police (west mountain between Gange & James)	8834	-001	24	76.8	30.8
Site, North Side - Borehole #2	9246	-025	24	85.4	27.8
Firehall	9744	-010	24	68.9	26.7
Blessed Kateri Tekakwitha School	9268	-002	24	50.9	17.2

TABLE II AIR PARTICULATE ANALYSIS: REVERTANTS PER PLATE
 Site: Mountain Police
 High Vol #8834
 Filter #29096001

Experiment #261082

#271082

#281082

Dose µg/plate	m ³	TA 98			Dose µg/plate	m ³	TA 100			Dose µg/plate	m ³	TA 1537		
		-S9 Mean	SEM	+S9 Mean			-S9 Mean	SEM	+S9 Mean			-S9 Mean	SEM	+S9 Mean
1280	41.5	<u>40.0</u>	2.3	<u>130.3</u>	1280	41.5	<u>146.3</u>	6.7	<u>149.0</u>	1280	41.5	<u>55.3</u>	4.7	<u>41.0</u>
960	31.1	<u>31.3</u>	1.9	<u>96.7</u>	960	31.1	<u>157.3</u>	14.8	<u>164.3</u>	960	31.1	<u>38.0</u>	3.8	<u>37.3</u>
640	20.8	<u>25.7</u>	5.4	<u>89.3</u>	640	20.8	<u>147.7</u>	2.3	<u>174**</u>	640	20.8	<u>43.0</u>	2.7	<u>29.0</u>
320	10.4	<u>28.0</u>	3.8	<u>57.3</u>	320	10.4	<u>132.7</u>	8.3	<u>129.0</u>	320	10.4	<u>39.3</u>	4.7	<u>21.7</u>
160	5.2	<u>19.3</u>	3.9	<u>32.7</u>	160	5.2	<u>141.7</u>	7.5	<u>139.3</u>					
0.1ml DMSO		<u>14.7</u>	3.3	<u>24.0</u>	0.1ml DMSO		<u>105.3</u>	3.3	<u>134.7</u>	0.1ml DMSO		<u>31.7</u>	1.2	<u>18.7</u>
0.5µg 4-NQO		386.0	19.7	-	0.5µg 4-NQO		>1000		6.4	150µg 9AA		230.7	24.3	-
10µg 2-AA		-		>1000	10µg 2-AA		-		>1000	10µg 2-AA		-		165.0
S9 only(0.2ml)		-		0	S9 only(0.2ml)		-		0	S9 only(0.2ml)		-		0

** only 1 plate scored

* " 2 plates "

TABLE III AIR PARTICULATE ANALYSIS: REVERTANTS PER PLATE
Site: Blessed Kateri Tekakwitha School
High Vol #9268
Filter #29096002

Experiment	#181082	#131082	#201082													
Dose μg/plate	m ³ -S9 Mean	TA 98 SEM	+S9 Mean	Dose μg/plate	m ³ -S9 Mean	TA 100 SEM	+S9 Mean	SEM	Dose μg/plate	m ³ -S9 Mean	TA 1537 +S9 Mean	SEM				
848	49.2	32.7	7.2	23.0	5.3	509	29.5	125**	131.7	14.7	848	49.2	23.0*	9.0	21.3	5.0
636	36.9	41.0	6.5	65.0	3.8	383	22.2	108.5*	135.0	15.3	636	36.9	19.3	5.0	20.0	2.9
424	24.6	35.7	3.3	43.0	2.1	255	14.8	107.7	115.3	0.9	424	24.6	15.3	3.5	14.0	2.0
212	12.3	19.7	2.3	33.0	5.8	128	7.4	95.0	98.0	3.6	212	12.3	13.7	2.2	14.0	1.0
106	6.2	29.7	2.4	28.0	4.0	64	3.7	92.2	94.7	5.0						
0.1ml DMSO		26.7	0.6	23.3	4.3	0.1ml DMSO		82.0	80.7	4.6	0.1ml DMSO		23.0	1.0	11.3	2.3
0.5μg 4-NQO	475.3	81.0	-	-		0.5μg 4-NQO		484.0	-		150μg 2-AA	184.0	66.0		-	
10μg 2-AA	-		>1000			10μg 2-AA		-	>1000		10μg 2-AA		-		146.7	6.4
S9 only(0.2ml)	-		0			S9 only(0.2ml)		-	0		S9 only(0.2ml)		-		0	
						#021182										
						471	27.3	134.3	139.3	7.9						
						236	13.7	140.5	120.7	7.8						
						0.1ml DMSO		116.7	122.3	6.7						
						2μg MNNG		>1000	-							
						S9 only(0.2ml)		-	0							

TABLE IV AIR PARTICULATE ANALYSIS: REVERTANTS PER PLATE
Site: Bell Canada Building
High Vol #9892
Filter #29096004

Experiment #081082

#081082

#161082

Dose µg/plate	m ³	-S9 Mean	+S9 Mean	Dose µg/plate	m ³	-S9 Mean	+S9 Mean	Dose µg/plate	m ³	-S9 Mean	+S9 Mean
7129	26.6	26.7	27.7	7129	26.6	111.3	110.0	1618	44.2	18.0	11.0
486	20.0	27.0	37.7	486	20.0	112.7	116.7	1214	33.1	15.7	12.7
243	13.3	18.3	22.0	243	13.3	119.0	113.3	809	22.1	19.0	13.0
121	6.7	16.3	21.0*	121	6.7	107.5	110.3	405	11.0	10.7	10.0
0.1ml DMSO	3.4	18.0	24.7	0.1ml DMSO	3.4	116.3	113.7	203	6.0	12.5	10.3
0.5 µg 4-NQO	15.3	2.0	23.3	0.5 µg MNNG	116.3	8.2	117.0	150 µg 9-AA	15.3	1.9	12.3
S9 only(0.2ml)	572.0	69.0	-	>1000	>1000	-	-	10 µg 2-AA	59.3	25.1	-
10 µg 2-AA	-	-	>1000	10 µg 2-AA	-	-	-	S9 only (0.2ml)	-	-	202.0
#021182	39.2	20.3	25.7	#021182	24.5	135.3	125.3	21.1	21.1	21.1	21.1
719	19.6	16.5	21.3	719	12.3	130.5	126.5				
0.1ml DMSO	13.3	2.9	22.0	0.1ml DMSO	116.7	13.9	122.3				
0.5 µg 4-NQO	578.0	14.0	-	0.5 µg MNNG	>1000	-	-				
10 µg 2-AA	-	-	>1000	10 µg 2-AA	-	-	-				
S9 only	-	-	0	S9 only	-	-	0				

TABLE V AIR PARTICULATE ANALYSIS: REVERTANTS PER PLATE

[illegible]

TABLE VI AIR PARTICULATE ANALYSIS: REVERENDS PER PLATE
Site: Ministry of Environment Instrument Shop, Stoney Creek
High Vol #8895
Filter #P9096006

[illegible]

TABLE VII AIR PARTICULATE ANALYSIS: FEVERTANTS PER PLATE
 Site: South Side Landfill
 High Vol #9258
 Filter #29096007

Experiment	#091082						#151082/191082						#201082					
Dose µg/plate	m ³	TA 98 -S9 Mean	SEM	+S9 Mean	SEM	Dose µg/plate	m ³	TA 100 -S9 Mean	SEM	+S9 Mean	SEM	Dose µg/plate	m ³	TA 1537 -S9 Mean	SEM	+S9 Mean	SEM	
654	21.2	27.0	7.0	31.7	6.4	1090	35.3	112.7	9.2	144.0	17.1	1090	35.3	-	-	18.0	0.5	
491	15.9	21.0	1.5	27.0	3.0	818	26.5	109.3	6.6	53.0 ^(a)	1.2	818	26.5	-	-	21.0	1.2	
327	10.6	17.0*	0.0	22.7	2.2	545	17.6	108.3	4.4	136.3	14.4	545	17.6	-	-	18.0	3.1	
164	5.3	13.3	2.2	18.7	0.9	273	8.8	126.0	5.0	145.3	11.7	273	8.8	-	-	14.0	2.1	
82	2.6	13.0	2.7	22.0	3.6	136	4.4	115.7	8.8	151.0	5.5							
0.1ml DMSO		13.7	2.3	19.3	3.0	0.1ml DMSO		114.3	0.9	133.3	3.7	C.2ml DMSO		-	-	11.3	2.3	
0.5µg 4-NQO		595.3	20.3	-	-	2µg MNNG		>1000		-		150µg 9-AA		184.0	66.0	-		
10µg 2-AA		-	-	>1000	-	10µg 2-AA		-	>1000	-		10µg 2-AA		-	-	146.7	6.4	
S9 only(0.2ml)		-	-	0	-	S9 only (0.2ml)		-	0	0	-	S9 only(0.2ml)		-	-	0	-	
#021182																		
848	27.5	19.5*	0.5	52.0*	2.0													
424	13.7	19.0*	3.0	41.3	4.7													
0.1ml DMSO		13.3	2.9	22.0	2.5													
0.5µg 4-NQO		578.0	14.0	-	-													
10µg 2-AA		-	-	>1000	-													
S9 only (0.2ml)		-	-	0	-													
(a) Toxicity Observed																		

(a) Toxicity Observed

TABLE VIII AIR PARTICULATE ANALYSIS: REVERTANTS PER PLATE
 Site: E.Side Landfill
 High Vol #8938
 Filter #2906008

Experiment	#261082	#271082	#281082														
Dose $\mu\text{g}/\text{plate}$	m^3 -S9 Mean	TA 98 SEM	+S9 Mean	SEM	Dose $\mu\text{g}/\text{plate}$	m^3 -S9 Mean	TA 100 SEM	+S9 Mean	SEM	Dose $\mu\text{g}/\text{plate}$	m^3 -S9 Mean	TA 1537 SEM	+S9 Mean	SEM			
1825	43.9	48.3	6.01	152.7	9.7	1825	43.9	123.0	2.5	156.0	8.5	1825	43.9	53.7	3.5	39.7	2.9
1369	32.9	36.0	4.0	75.3	8.1	1369	32.9	140.3	7.4	161.7	16.7	1369	32.9	40.0	2.7	31.3	1.7
913	22	30.7	0.7	67.3	3.7	913	22	147.3	9.3	139.3	10.5	913	22	34.0	5.0	22.0	4.9
456	11	26.7	2.8	36.3	3.3	456	11	153.7	10.2	146.3	13.0	456	11	18.0	2.1	12.7	4.3
228	5.5	17.3	1.9	30.3	4.1	228	5.5	143.3	2.4	140.3	6.6						
0.1ml DMSO		14.7	3.3	24.0	2.1	0.1ml DMSO		105.3	3.3	134.7	6.4	0.1ml DMSO		31.7	1.2	18.7	2.4
0.5 μg 4-NQO		386.0	19.7	-		0.5 μg 4-NQO		>1000		-		150 μg 9-AA		230.7	24.3	-	
10 μg 2-AA		-		>1000		10 μg 2-AA		-		>1000		10 μg 2-AA		-		165.0	9.3
S9 only(0.2ml)		-		0		S9 only(0.2ml)		-		0		S9 only (0.2ml)		-		0	

TABLE IX AIR PARTICULATE ANALYSIS: REVERTANTS PER PLATE
 Site: South East Side Landfill
 High Vol #9495
 Filter #29096009

Experiment		#261082				#271082				#281082			
Dose µg/plate	m ³	TA 98		+S9		Dose µg/plate	m ³	TA 100		Dose µg/plate	m ³	TA 1537	
		-S9 Mean	SEM	Mean	SEM			-S9 Mean	SEM			+S9 Mean	SEM
1688	45.4	50.7	9.7	153.7	12.8	1688	45.4	167.0	8.7	169.0	6.8	168.8	45.4
1266	34.0	36.3	5.4	120.0	6.7	1266	34.0	150.3	8.7	174.3	8.9	1266	34.0
844	22.7	38.7	2.4	78.7	13.0	844	22.7	137.7	9.7	160.0	4.6	844	22.7
422	11.3	25.0	0.6	-	-	422	11.3	134.3	4.4	131.0	4.6	422	11.3
						211	5.7	138.3	4.5	118.0	6.1	0.1ml DMSO	230.7
0.1ml DMSO		14.7	3.3	24.0	2.1	0.2ml DMSO		105.3	3.3	134.7	6.4	150µg 9-AA	24.3
0.5µg 4-NQO		386.0	19.7	-	-	0.5µg 4-NQO		>1000	-	-	-	-	-
10µg 2-AA		-	-	>1000	-	10µg 2-AA		-	-	>1000	-	10µg 2-AA	-
S9 only(0.2ml)		-	-	0	-	S9 only (0.2ml)		0	-	-	-	0	-

TABLE X AIR PARTICULATE ANALYSIS: REVERTANTS PER PLATE

Site: Firehall
High Vol #9744
Filter #29096010

Experiment		#111082		#191082		#211082	
		TA 98		TA 100		TA 1537	
Dose $\mu\text{g}/\text{plate}$	m^3	-S9 Mean	SEM	+S9 Mean	SEM	-S9 Mean	SEM
Dose $\mu\text{g}/\text{plate}$	m^3	-S9 Mean	SEM	+S9 Mean	SEM	-S9 Mean	SEM
689	25.8	38.0	3.0	58.0	0.6	1148	42.9
517	19.3	32.7	2.9	36.0	1.5	861	32.2
345	12.9	33.0	1.5	36.3	1.2	574	21.5
172	6.5	21.7	2.4	23.3	1.3	287	10.7
86	3.2	19.3	1.8	26.7	1.9	144	5.4
0.1ml DMSO		22.0	2.7	27.3	5.8	0.1ml DMSO	
0.5 μg 4-NQO		557.7	42.0	-		0.5 μg 4-NQO	
10 μg 2-AA		-		>1000		10 μg 2-AA	
S9 only (0.2ml)		-		0		S9 only (0.2ml)	
#021782							
1020	38.2	25.0	2.0	69.3	7.1		
510	19.1	24.0	1.0	47.3	4.3		
0.1ml DMSO		13.3	2.9	22.0	2.5		
0.5 μg 4-NQO		578.0	14.0	-			
10 μg 2-AA		-		>1000			
S9 only (0.2ml)		-		0			
#191082							
689	25.8	38.0	3.0	138.7	8.1	1148	42.9
517	19.3	32.7	2.9	115.3	14.8	861	32.2
345	12.9	33.0	1.5	133.7	8.6	574	21.5
172	6.5	21.7	2.4	145.3	3.2	287	10.7
86	3.2	19.3	1.8	117.0	10.5	144	5.4
0.1ml DMSO		22.0	2.7	119.7	2.6	0.1ml DMSO	
0.5 μg 4-NQO		557.7	42.0	>1000		0.5 μg 4-NQO	
10 μg 2-AA		-		-		10 μg 2-AA	
S9 only (0.2ml)		-		0		S9 only (0.2ml)	
#211082							
689	25.8	38.0	3.0	141.3	13.1	1148	42.9
517	19.3	32.7	2.9	149.7	2.6	861	32.2
345	12.9	33.0	1.5	151.7	9.7	574	21.5
172	6.5	21.7	2.4	136.0	3.6	287	10.7
86	3.2	19.3	1.8	140.0	15.7		
0.1ml DMSO		22.0	2.7	133.3	3.7	0.1ml DMSO	
0.5 μg 4-NQO		557.7	42.0	-		150 μg 9-AA	
10 μg 2-AA		-		>1000		10 μg 2-AA	
S9 only (0.2ml)		-		0		S9 only (0.2ml)	
#211082							
689	25.8	38.0	3.0	23.7	3.8	1148	42.9
517	19.3	32.7	2.9	28.7	1.8	861	32.2
345	12.9	33.0	1.5	14.0	2.5	574	21.5
172	6.5	21.7	2.4	25.3	1.2	287	10.7
86	3.2	19.3	1.8				
0.1ml DMSO		22.0	2.7	15.3	0.88	0.1ml DMSO	
0.5 μg 4-NQO		557.7	42.0	228.7	25.3	150 μg 9-AA	
10 μg 2-AA		-		-		10 μg 2-AA	
S9 only (0.2ml)		-		-		S9 only (0.2ml)	
#211082							
689	25.8	38.0	3.0	26.7	1.3	1148	42.9
517	19.3	32.7	2.9	26.3	4.3	861	32.2
345	12.9	33.0	1.5	19.0	2.5	574	21.5
172	6.5	21.7	2.4	19.0	3.6	287	10.7
86	3.2	19.3	1.8				
0.1ml DMSO		22.0	2.7	17.3	1.9	0.1ml DMSO	
0.5 μg 4-NQO		557.7	42.0	-		150 μg 9-AA	
10 μg 2-AA		-		181.0	16.6	10 μg 2-AA	
S9 only (0.2ml)		-		1		S9 only (0.2ml)	

Site: North Gate, Landfill
High Vol #9246
Filter #29096025

Experiment	#181082					#131082					#211082				
	TA 98					TA 100					TA 1537				
Dose	m ³	-S9	SEM	+S9	Dose	m ³	-S9	SEM	+S9	Dose	m ³	-S9	SEM	+S9	
µg/plate	Mean	Mean	Mean	Mean	µg/plate	Mean	Mean	Mean	Mean	µg/plate	Mean	Mean	Mean	Mean	
1423	51.1	<u>47.0</u>	3.8	<u>71.3</u>	854	30.7	<u>118.5*</u>	5.5	<u>272.5*</u>	1423	51.1	<u>23.3</u>	2.2	<u>24.0</u>	
1068	38.4	<u>37.0</u>	2.1	<u>60.0</u>	641	23.0	<u>113.0*</u>	11.0	<u>248.0*</u>	1068	38.4	<u>24.3</u>	3.5	<u>35.3</u>	
712	25.6	<u>37.7</u>	2.2	<u>52.0</u>	427	15.3	<u>110.0</u>	3.8	<u>148.3</u>	712	25.6	<u>21.7</u>	1.2	<u>25.3</u>	
356	12.8	<u>31.7</u>	2.2	<u>49.0</u>	214	7.7	<u>97.0</u>	3.8	<u>99.7</u>	356	12.8	<u>18.3</u>	2.4	<u>16.0</u>	
178	6.4	<u>25.0</u>	0.6	<u>31.3</u>	107	3.8	<u>91.7*</u>	10.1	<u>109.0</u>						
0.1ml DMSO		<u>26.7</u>	0.6	<u>23.3</u>			<u>82.0</u>	6.0	<u>80.7</u>	0.1ml DMSO		<u>15.3</u>	0.9	<u>17.3</u>	
0.5µg 4-NQO		475.3	81.0	-	0.5µg 4-NQO		484.0	35.9	-	150µg 9-AA		228.7	25.3	-	
10µg 2-AA		-		>1000	10µg 2-AA		-		>1000	10µg 2-AA		-		181.0	
S9 only(0.2ml)		-		0	S9 only (0.2ml)		-		0	S9 only(0.2ml)		-		0	

TABLE XII MUTAGENICITY VALUES FROM REGRESSION ANALYSIS FOR TA 98

Location	Revertants/mg		Revertants/m ³		Correlation Coefficient	
	-S9	+S9	-S9	+S9	-S9	+S9
Mountain Police (control)	17	82	.5	2.5	.94	.99
Blessed Kateri Tekakwitha School	25	80	.4	1.4	.77	.97
Bell Canada Building	14	16	.5	.6	.86	.70
Benneto School (control)	29	43	.9	1.4	.97	.96
MOE Instrumentation Lab (control)	37	76	2.7	5.7	.98	.99
Site, S. Side	21	18	.7	.6	.96	.93
Site, E. Side	17	64	.7	2.7	.98	.94
Site, S. E. Side (Stonechurch Rd.)	20	77	.7	2.9	.96	1.0
Firehall	27	42	.7	1.1	.94	.88
Site, N. Side	14	31	.4	.9	.95	.96

TABLE XIII MUTAGENICITY VALUES FROM REGRESSION ANALYSIS FOR TA 100

Location	Revertants/mg		Revertants/m ³		Correlation Coefficient	
	-S9	+S9	-S9	+S9	-S9	+S9
Mountain Police (control)	43	41	1.3	1.3	.84	.79
Blessed KateriTekakwitha School	75	135	1.3	2.3	.97	.99
Bell Canada Building	-	-	-	-	-.28	-.34
Benneto School (control)	22	11	1.0	0.3	.86	.74
MOE Instrumentaion Lab. (control)	-	31	-	2.3	.25	.92
Site S.Side	-	-	-	-	-.44	.1
Site E.Side	-	16	-	1.0	.07	.81
Site S.E.Side (Stonechurch Rd.)	28	41	1.0	1.5	.89	.9
Firehall	-	21	-	1.0	.26	.9
Site N.Side	41	240	1.2	6.7	.96	.97

TABLE XIV MUTAGENICITY VALUES FROM REGRESSION ANALYSIS FOR TA 1537

Location	Revertants/mg		Revertants/m ³		Correlation Coefficient	
	-S9	+S9	-S9	+S9	-S9	+S9
Mountain Police (control)	7	19	.2	.6	.62	.99
Blessed Kateri Tekakwitha School	0	12	-	.2	.21	.96
Bell Canada Building	0	0	0	0	.59	.2
Benneto School (control)	0	11	0	.4	-.29	.89
MoE Instrumentation Lab (control)	9	16	.6	1.2	.99	.98
Site, S.Side	ND	12	ND	.4	ND	.99
Site, E.Side	15	12	.4	.3	.8	.94
Site, S.E. Side Stonechurch Rd.	0	15	0	.6	.5	.98
Firehall	10	9	.3	.2	.51	.92
Site N.Side	9	18	.2	.5	.99	.92

TABLE XV

CHARACTERISTICS OF LEACHATE SAMPLES

Site	pH	Characteristics
Upstream, Red Hill Creek	8.0	Clear, some sediment
Landfill, Borehole #2	8.4	Dark brown, oily, black sediment
Downstream, Red Hill Creek	8.3	Clear, some sediment
Hamilton Tap Water	7.8	Clear
Landfill, Collection Manhole	8.3	Dark brown, particles in suspension, green tinge.
Landfill, Borehole #4	8.2	Black, heavy sediment oily.

TABLE XVI LEACHATE ANALYSIS: REVERTANTS PER PLATE

Site Upstream Red Hill Creek

Experiment# 050882

100982

080982

170882

230982

Dose ml/plate	TA 98			TA 100			TA 1535			TA 1537			TA 1538		
	-S9 Mean	SEM	+S9 Mean	-S9 Mean	SEM	+S9 Mean	-S9 Mean	SEM	+S9 Mean	-S9 Mean	SEM	+S9 Mean	-S9 Mean	SEM	+S9 Mean
2.0	<u>21.3</u>	1.8	<u>31.0</u>	<u>113.0</u>	11.6	<u>107.3</u>	<u>31.3</u>	3.2	<u>16.3</u>	<u>2.4</u>	<u>14.0</u>	2.5	<u>10.7</u>	2.4	<u>2.0</u>
1.5	<u>17.7</u>	0.3	<u>34.0</u>	<u>106.3</u>	6.3	<u>100.0</u>	<u>27.3</u>	5.8	<u>16.3</u>	2.6	<u>9.7</u>	0.3	<u>11.7</u>	1.5	<u>1.3</u>
1.0	<u>19.7</u>	2.0	<u>38.3</u>	<u>119.3</u>	7.7	<u>130.7</u>	<u>33.3</u>	5.0	<u>13.3</u>	2.6	<u>19.7</u>	7.5	<u>17.7</u>	2.4	<u>3.7</u>
0.5	<u>21.7</u>	4.7	<u>31.3</u>	<u>133.3</u>	12.0	<u>125.7</u>	<u>37.0</u>	3.2	<u>11.3</u>	2.0	<u>13.3</u>	2.9	<u>11.7</u>	1.2	<u>2.0</u>
0.25	<u>20.7</u>	0.3	<u>28.7</u>	-	-	-	-	-	-	-	-	-	-	-	-
2ml deionized H ₂ O	<u>20.8</u>	1.3	<u>29.5*</u>	<u>118.3</u>	14.1	<u>117.5*</u>	<u>32.3</u>	1.3	<u>14.7</u>	0.3	<u>18.3</u>	3.8	<u>12.7</u>	3.3	<u>5.0</u>
0.5µg 4-NQO	596.0*	204.0*	-	-	-	-	-	-	-	-	-	-	-	-	-
150µg 9-AA	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
2µg MNNG	-	-	-	>1000	-	-	>1000	-	-	-	-	-	-	-	-
10µg 2-AA	-	-	>1000	-	-	>1000	-	-	214.3	18.8	-	-	173.0	33.0	-
S9 only	-	-	-	-	0	-	-	0	0	-	-	0	0	-	0
2ml Sample only	-	-	-	0	-	-	0	-	-	0	-	-	-	-	-

217.0

3.0

091182

Dose ml/plate	TA 98			TA 100			TA 1535			TA 1537			TA 1538				
	-S9 Mean	SEM	+S9 Mean	-S9 Mean	SEM	+S9 Mean	-S9 Mean	SEM	+S9 Mean	-S9 Mean	SEM	+S9 Mean	-S9 Mean	SEM	+S9 Mean	SEM	
2.0	<u>24.7</u>	1.2	<u>23.3</u>	<u>112.7</u>	5.6	<u>142.7</u>	<u>39.7</u>	4.7	<u>24.0</u>	2.7	<u>8.5*</u>	2.5	-ND-	<u>4.0</u>	1.0	<u>3.7</u>	1.9
1.5	<u>19.7</u>	3.5	<u>18.3</u>	<u>101.7</u>	12.7	<u>124.3</u>	<u>41.0</u>	2.5	<u>24.0</u>	1.2	<u>19.0</u>	5.6	<u>18.5</u>	<u>3.7</u>	1.5	<u>3.0</u>	1.5
1.0	<u>19.3</u>	2.9	<u>28.3</u>	<u>101.3</u>	9.4	<u>118.7</u>	<u>42.3</u>	0.7	<u>22.0</u>	1.2	<u>16.0</u>	5.1	<u>13.3</u>	<u>1.0</u>	0.6	<u>1.7</u>	0.3
0.5	<u>23.7</u>	1.8	<u>29.0</u>	<u>99.0</u>	8.3	<u>125.3</u>	<u>43.3</u>	4.3	<u>16.7</u>	4.2	<u>27.7</u>	4.1	<u>15.0</u>	<u>1.7</u>	0.3	<u>2.0</u>	0.6
0.25	<u>23.0</u>	2.9	<u>33.0</u>	-	-	-	-	-	-	-	-	-	-	-	-	-	-
2ml deionized H ₂ O	<u>21.7</u>	1.2	<u>29.0</u>	<u>108.0</u>	4.2	<u>114.8</u>	<u>49.0</u>	6.7	<u>15.3</u>	0.3	<u>21.3</u>	4.1	<u>15.8</u>	<u>2.0</u>	1.0	<u>3.0</u>	1.0
0.5µg 4-NQO	560.0	56.8	-	-	-	-	-	-	-	-	-	-	-	55.9	7.9	-	-
2µg MMS	-	-	-	>1000	-	-	>1000	-	-	-	-	-	-	-	-	-	-
150µg 9-AA	-	-	-	-	-	-	-	-	-	-	105.0*	27.0	-	-	-	-	-
10µg 2-AA	-	>1000	-	-	>1000	-	-	217.7	18.2	-	-	284.0	30.3	-	-	TOX	-
S9 only	-	-	-	-	0	-	-	ND	-	-	-	0	-	-	0	-	-
Sample only	-	-	-	0	-	-	0	0	-	0	0	-	-	0	-	-	-

TABLE XVIII LEACHATE ANALYSIS: REVERTANTS PER PLATE
Site Downstream, Red Hill Creek
Experiment 050882 100982 080982 170882 230982

Dose ml/plate	TA 98			TA 100			TA 1535			TA 1537			TA 1538		
	-S9 Mean	SEM	+S9 Mean	-S9 Mean	SEM	+S9 Mean	-S9 Mean	SEM	+S9 Mean	-S9 Mean	SEM	+S9 Mean	-S9 Mean	SEM	+S9 Mean
2.0	<u>22.3</u>	1.5	<u>34.3</u>	<u>137.0</u>	4.2	<u>155.0</u>	<u>34.3</u>	4.8	<u>15.0</u>	<u>12.0</u>	2.7	<u>10.3</u>	<u>4.3</u>	0.9	<u>4.0</u>
1.5	<u>24.0</u>	0.0	<u>27.7</u>	<u>154.0</u>	10.1	<u>138.3</u>	<u>31.3</u>	2.3	<u>15.0</u>	<u>7.3</u>	0.9	<u>11.0</u>	<u>3.3</u>	0.7	<u>3.0</u>
1.0	<u>22.0</u>	1.5	<u>32.0</u>	<u>164.0</u>	10.8	<u>128.0</u>	<u>35.5*</u>	3.5	<u>17.3</u>	<u>8.7</u>	2.7	<u>19.0</u>	<u>5.7</u>	0.7	<u>1.3</u>
0.5	<u>24.0</u>	1.0	<u>36.0</u>	<u>168.0</u>	34.0	<u>132.3</u>	<u>31.0</u>	2.9	<u>16.3</u>	<u>8.0</u>	2.1	<u>18.0</u>	<u>3.3</u>	0.3	<u>3.0</u>
0.25	<u>25.7</u>	3.5	<u>28.3</u>	-	-	-	-	-	-	-	-	-	-	-	-
2ml deionized H ₂ O	<u>20.8</u>	1.3	<u>29.5*</u>	<u>118.3</u>	14.0	<u>117.5*</u>	<u>32.3</u>	1.3	<u>14.7</u>	<u>18.3</u>	3.8	<u>12.7</u>	<u>5.0</u>	1.7	<u>2.7</u>
0.5µg 4-NQO	596.0	204.0	-	-	-	-	-	-	-	-	-	-	-	-	-
2µg MNNG	-	-	-	>1000	-	-	>1000	-	-	-	-	-	-	-	-
150µg 9AA	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
10µg 2-AA	-	-	>1000	-	-	>1000	-	-	214.3	18.8	-	173.0*33.0	-	-	217.0*
Sample only	-	-	-	0	-	-	0	-	-	0	-	-	-	-	-
S9 only	-	-	-	-	0	-	-	0	-	-	-	0	-	-	0

TABLE XIX LEACHATE ANALYSIS: REVERTANTS PER PLATE
 Site Hamilton Taw Water
 Experiment# 050882 100982 080982 300982 230982

Dose ml/plate	TA 98			TA 100			TA 1535			TA 1537			TA 1538		
	-S9 Mean	SEM	+S9 Mean	-S9 Mean	SEM	+S9 Mean	-S9 Mean	SEM	+S9 Mean	-S9 Mean	SEM	+S9 Mean	-S9 Mean	SEM	+S9 Mean
2.0	<u>23.7</u>	2.6	<u>33.0</u>	<u>133.7</u>	6.5	<u>115.3</u>	<u>13.4</u>	<u>43.0</u>	<u>17.3</u>	<u>15.3</u>	1.7	<u>21.0</u>	<u>3.3</u>	1.2	<u>3.7</u>
1.5	<u>19.0</u>	3.5	<u>33.3</u>	<u>124.0</u>	6.2	<u>107.3</u>	7.6	<u>22.0</u>	<u>18.0</u>	<u>19.0</u>	4.5	<u>20.5*</u>	<u>2.0</u>	0.6	<u>2.0</u>
1.0	<u>18.0</u>	1.0	<u>20.7</u>	<u>133.3</u>	6.9	<u>137.7</u>	14.1	<u>25.3</u>	<u>29.7</u>	<u>17.7</u>	5.0	<u>21.0</u>	<u>2.7</u>	1.2	<u>1.3</u>
0.5	<u>16.0</u>	2.1	<u>26.7</u>	<u>142.7</u>	9.9	<u>136.5*</u>	2.5	<u>33.3</u>	<u>16.7</u>	<u>23.0</u>	4.2	<u>15.7</u>	<u>2.5</u>	0.5	<u>1.3</u>
0.25	<u>20.7</u>	6.8	-	-	-	-	-	-	-	-	-	-	-	-	-
2ml deionized H ₂ O	<u>20.8</u>	1.3	<u>29.5*</u>	<u>118.3</u>	14.1	<u>117.5</u>	14.5	<u>32.3</u>	<u>14.7</u>	<u>21.3</u>	4.1	<u>15.8</u>	<u>5.0</u>	1.7	<u>2.7</u>
0.5µg 4-NQO	596.0*204.0*	-	-	-	-	-	-	-	-	-	-	-	-	-	-
2µg MNNG	-	-	-	>1000	-	-	>1000	-	-	-	-	-	-	-	-
150 µg 9-AA	-	-	-	-	-	-	-	-	-	105.0* 27.0	-	-	-	-	-
10µg 2-AA	-	>1000	-	-	>1000	-	-	-	214.3 18.8	-	284.0 30.3	-	-	217.0*	3.0
Sample only	-	-	-	0	-	-	0	-	-	0	-	-	-	-	-
S9 only	-	-	-	-	-	-	-	-	0	-	0	-	-	0	-

TABLE XX LEACHATE ANALYSIS: REVERTANTS PER PLATE
Site Collection Manhole
Experiment # 060822

200982

090982

300982

230952

Dose ml/plate	TA 98			TA 100			TA 1535			TA 1537			TA 1538		
	-S9 Mean	SEM	+S9 Mean	-S9 Mean	SEM	+S9 Mean	-S9 Mean	SEM	+S9 Mean	-S9 Mean	SEM	+S9 Mean	-S9 Mean	SEM	+S9 Mean
2.0	<u>21.7</u>	5.2	<u>27.0</u>	<u>27.7</u>	6.8	<u>116.7</u>	<u>45.7</u>	3.5	<u>24.3</u>	<u>26.0*</u>	1.0	<u>13.5*</u>	<u>3.3</u>	0.9	<u>4.0</u>
1.5	<u>19.0</u>	1.2	<u>27.7</u>	<u>88.3</u>	4.4	<u>110.7</u>	<u>43.0</u>	2.5	<u>20.0</u>	<u>18.5*</u>	2.5	<u>14.5</u>	<u>2.3</u>	0.7	<u>3.5*</u>
1.0	<u>24.7</u>	3.8	<u>34.0</u>	<u>90.3</u>	2.9	<u>112.7</u>	<u>50.3</u>	8.4	<u>26.5*</u>	<u>22.5*</u>	4.5	<u>17.0*</u>	<u>2.7</u>	1.7	<u>2.3</u>
0.5	<u>32.0</u>	2.0	<u>30.7</u>	<u>101.0</u>	4.2	<u>118.7</u>	<u>47.3</u>	4.8	<u>18.0</u>	<u>19.3</u>	4.8	TOX	<u>2.0</u>	0.6	<u>3.5</u>
0.25	<u>20.0</u>	1.0	<u>36.3</u>	-	-	-	-	-	-	-	-	-	-	-	-
2ml deionized H ₂ O	<u>21.7</u>	1.2	<u>29.0</u>	<u>108.0</u>	4.2	<u>114.8</u>	<u>49.0</u>	6.7	<u>15.3</u>	<u>21.3</u>	4.1	<u>15.8</u>	<u>5.0</u>	1.7	<u>2.7</u>
C. 5µg 4-NQO	560.0	56.8	-	-	-	-	-	-	-	-	-	-	-	-	-
2µg MINT?	-	-	-	>1000	-	-	>1000	-	-	-	-	-	-	-	-
150µg 9-AA	-	-	-	-	-	-	-	-	-	105.0*	27.0	-	-	-	-
10µg 2-AA	-	>1000	-	-	>1000	-	-	217.7	18.2	-	284.0	30.3	-	217.0	3.0
Sample only	-	-	-	0	-	-	0	-	-	0	-	-	-	-	-
S9 only	-	-	-	-	0	0	-	-	-	-	0	0	-	-	0

STRAIN CHECK

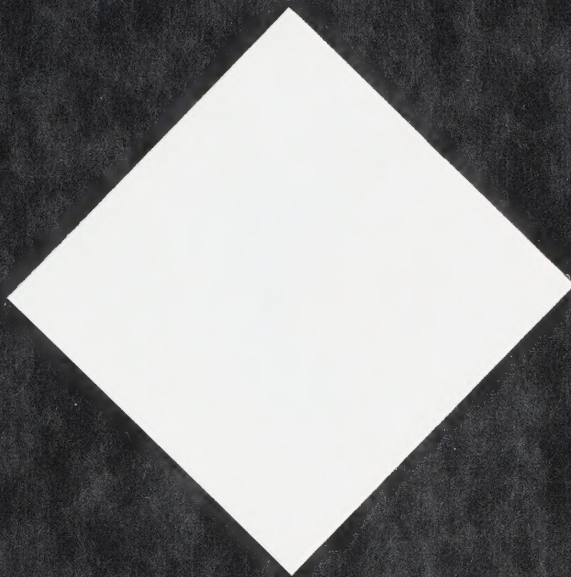
DATE 8th Oct 82.
STOCK Setup 7 Oct from
stocks frozen 7 Oct.

STRAIN	TA98	TA100	TA1535	TA1537	TA1538
GROWTH IN PRESENCE OF HIS	✓	✓	✓	✓	✓
GROWTH IN ABSENCE OF HIS	✓	✓	✓	✓	✓
CRYSTAL VIOLET: ZONE OF INHIBITION	✓	✓	✓	✓	✓
UV, 6SEC			✓	✓	✓
GOWTH ON ONLY HALF PLATE					
UV, 8SEC	✓	✓			
AMPICILLIN INHIBITION	✓	✓	✓	✓	✓
SPONT REVERSION:	12	110	19	4	12
			29	0	10
SPOT TEST					
INDUCED REVERSION:					
MNNG	✓	✓	✓	✓	✓
4NQO	✓	✓	✓	✓	✓
9AA	✓	✓	✓	✓	✓

11 Oct 82.

16/10 1537 vs 150µg/plate 9AA - clear +ve





ACCOPRESS®

25071	—	BLACK / NOIR	—	BG2507
25072	—	BLUE / BLEU	—	BU2507
25078	—	RED / ROUGE	—	BF2507
25075	—	GREEN / VERT	—	BP2507
25074	—	GREY / GRIS	—	BD2507
25073	—	R. BLUE / BLEU R.	—	BB2507
25079	—	X. RED / ROUGE X.	—	BX2507
25070	—	YELLOW / JAUNE	—	BY2507
25077	—	TANGERINE	—	BA2507

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